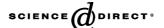


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Effect of high-pressure hot water washing treatment on fruit quality, insects, and disease in apples and pears Part IV: Use of silicone-based materials and mechanical methods to eliminate surface arthropod eggs

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Abstract

The presence of surface arthropods on commercially processed apples and pears poses a problem when these fruits are exported to countries where there are either limits on the numbers of eggs or a total quarantine restriction against these pests. Removal of mite and other arthropod eggs, such as European red mite (ERM) and codling moth (CM) eggs, may be enhanced by the use of a surface cleaning system, such as a hot water, high-pressure spray. Even if organosilicones, like Silwet L-77, have been used to kill spider mites, it was unclear if these chemicals could also facilitate the removal of arthropod eggs from the surface of fruit. In the present study, high-pressure washing process was highly effective in removing CM and ERM eggs at pressures as low as 400 kPa, resulting in greater than 60% removal of ERM eggs and 90% of CM eggs. High-pressure washing was the most important factor in removal of codling moth and European red mite eggs than organosilicone dips or hot water sprays.

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Keywords: Pyrus communis; Cydia pomonella; Panoychus ulmi; High-pressure washing; Organosilicone; Postharvest phytosanitation

1. Introduction

The presence of lepidopteran and European red mite (ERM) *Panonychus ulmi* (Koch) eggs on pears has been a concern for many export markets. Typically, the removal of these eggs has proved to be problematic. Therefore, cold storage has been recommended for control of the lepidopteran eggs, whereas, a tolerance of only 5% of the fruit infested with ERM eggs has been set for many countries (NWHC, 2004). A hot water spray system could either remove the eggs or kill the embryos due to either the pressure of the wash or the temperature of the spray.

High-pressure washing using hot water was developed in Israel to improve commodity postharvest quality and reduce decay (Akerman, 1997; Fallik et al., 1999, 2000a,b; Lichter et al., 2000; Porat et al., 2000; Prusky et al., 1999). The system was originally designed to clean and disinfest fruit of insects and rots. Numerous reports indicated that hot water sprays of 40–55 °C effectively controlled decay organisms and many arthropods (Akerman, 1997; Fallik et al., 1999, 2000a,b; Karabulut et al., 2002; Porat et al., 2000; Prusky et al., 1999).

Silwet L-77, an organosilicone surfactant and common spray adjuvant (Helena Chemical Company, Memphis, TN), has been shown to be an effective miticide (Cowles et al., 2000; Dentener and Peetz, 1992; Liu, 2000; Purcell and Schroeder, 1996; Tipping et al., 2003; Wood, 1997), and may

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also control some fungi (Blenis, 1997). The mode of action of Silwet is believed to be through the blocking of the spiracles, causing suffocation of the mites (Cowles et al., 2000). However, dead mites still cause a problem for many importing countries.

In the Pacific Northwest, high-pressure washing became popular after the advent of kaolin clay applications for tree fruit (Glenn et al., 2001; Knight et al., 2000; Puterka and Glenn, 2000; Unruh et al., 2000). This clay is highly hydrophobic and difficult to clean off the fruit with existing dip tanks, soapers, and bushes. High-pressure sprays of 400-700 kPa became popular as a cleaning method. However, initial surveys of the pear industry indicated that decay organisms were being spread throughout the load. The high-pressure washing system uses recirculating water, which was largely untreated and ended up being a source of decay microorganisms. In this study, we included a heated contact loop to control decay organisms and added a counter current heat exchanger to allow for finer control of spray water temperatures (Bai et al., 2006). This system allowed us to investigate the impact of high-pressure washing in conjunction with hot water for removal of surface arthropods.

The objective of this study was to evaluate the removal of lepidopteran eggs and ERM eggs from apples and pears using different concentrations of Silwet, followed by water spraying at different temperatures and pressures. Our interest in Silwet was as a surfactant and its ability to penetrate the hydrophobic calyx end of pome fruit, where spider mites, mealy bugs, and a plethora of other small arthropods reside. In Washington State, Silwet is classified as a pesticide because it is used as a pesticide spray adjuvant. This classification causes significant concerns over the disposal of solutions containing this agent. Therefore, other surfactants more acceptable to regulatory and food agencies (i.e. those having food grade registration) were also investigated for their ability to penetrate the calyx and facilitate the removal of surface arthropods.

2. Materials and methods

2.1. Experimental design

Four tests were conducted between December 2001 and April 2004 to determine removal of various targeted surface arthropod eggs on an experimental packing line at the Oregon State University Experiment Station in Hood River, Oregon. The eggs of codling moth, *Cydia pomonella* (L.) and oriental fruit moth, *Grapholitha molesta* (Busck) were used in Test 1. The eggs of European red mite, *Panonychus ulmi* (Koch), and codling moth were used in Test 2, while only the mature eggs of codling moth were tested in Tests 3 and 4. Except for Test 1, all infested fruit were tested on the experimental packing line. The packing line contains a high-pressure hot water system which consists of a boiler, hot water mixing tank, contact

loop, heat exchanger, high-pressure pump, spray tank, highpressure spray manifold, and low-pressure fresh-water spray manifold (Bai et al., 2006). Treatments used in Tests 2-4 used various combinations of three water spray temperatures (25, 40, and 50 °C), six spray pressures (0, 200, 400, 550, 700, and 850 kPa), three concentrations of Silwet (0, 0.2, and 0.3%), three concentrations of silicone defoamer (0, 0.01, and 0.1%), (Ivanhoe Industries Inc., Mundelein, IL), and two styles of washing brushes, 0.38 mm (hard) or 0.30 mm (soft) PEC bristles (American Brush Company, Portland, OR) were tested. Chemical dips were done using large tubs holding 20 L of water in which fruit were held for 60 s. Then the fruit were placed onto the packing line, just after the dump tank, run through the water spray system, and removed just before the dryer section. After all treatments were completed, the fruit were put in holding containers and returned to the Wapato laboratory for evaluation.

2.1.1. Lepidoptera eggs

Mature, organic 'Red Delicious' apples were washed with 100 ppm of chlorine and hand dried with cotton towels. Fruit at 20 °C were placed as a single layer into cardboard boxes $(762 \text{ mm} \times 762 \text{ mm})$ lined with plastic sheets. Codling moths (500 total, 250 female, and 250 male) or oriental fruit moths, were briefly chilled and placed on top of apples in the box. A lid covered with nylon mesh was placed over the box of apples and moths. The box was then placed into an environmental chamber with a photoperiod of 16-h light/8-h dark, at 23 °C, and 50–60% RH. Moths were allowed to oviposit on the fruit for 24 h. The box was then placed into a cold room (0 °C) for 600 s, after which the moths were vacuumed off the fruit and out of the box. Fruit were examined for eggs using a dissecting microscope. Each apple was numbered and the number of eggs per fruit recorded. Fruit were divided into groups to provide a total of 200 eggs per treatment group. Mature eggs, called black head, were used for the tests since they were the most resistant to crushing and high water temperatures.

2.1.2. European red mites

Diapausing eggs of European red mite were obtained from naturally infested fruit from a commercial packing house in Yakima, WA. Fruit were inspected for ERM eggs with a dissecting microscope. The number of eggs per fruit was determined and marked on the fruit with a waterproof marker.

2.2. Test 1

A preliminary test to examine the impact of Silwet L-77 on egg hatch alone was performed on the eggs of codling moth and oriental fruit moth. Fruit were infested as previously described, the only difference being that white ring (0–3 d) and black head (5–7 d) stages were used. A solution of 0.75% Silwet L-77 in deionized water was used to dip Red Delicious apples with the moth eggs on them. Dips were carried out for 0, 60, 300, and 600 s. The fruit were removed from the solution and placed in plastic deli containers and set into a

rearing room (16-h light/8-h dark, 23 °C, 50–60% RH) for 7–14 d. Fruit were examined under a dissecting microscope after 7 and 14 d for egg hatch.

2.3. Test 2

The focus of this test was to determine the effects of water spray temperature, spray pressure, and Silwet concentration on the removal of ERM and mature codling moth eggs from the fruit surface. Treatments (Table 2) were a combination of the concentration of Silwet (0, 0.2, and 0.3%), hot water spray temperature (20 [control], 40, and 50 °C), and pressure (none, 200, and 550 kPa). Pears were added at the start (dump tank) of a simulated packing line. Control fruit were placed in a tub of water at room temperature (20 °C) for approximately 480 s, to simulate fruit in the packing line. The study was done in conjunction with other tests involving removal of surface arthropods, occurrence of disease, and fruit quality. After fruit went through the system (about 480 s), they were returned to the laboratory and the number of eggs per fruit were counted using a binocular dissecting microscope. Fruit were held at 16-h light/8-h dark, 23 °C, and 50–60% RH in an environmental room for 1–7 d prior to evaluation for removal and egg hatch, respectively. Fruit were examined for presence or absence of eggs for both ERM and CM and the number of remaining eggs that hatched for CM.

2.4. Test 3

The foci of this test was to determine the effects of water spray temperature, spray pressure, brush type, and defoamer concentration on the removal of CM eggs from the fruit surface. Treatments consisted of a combination of four factors: water spray temperature (10 or 27 °C), spray pressure (0 or 400 kPa), concentration of silicone defoamer (0.01 or 0.1%), and brush type (hard or soft). Brushes were 124 mm in diameter; with either 0.38 mm (hard) (Type A) or 0.30 mm (soft) (Type B) PEC bristles (American Brush Company, Portland, OR). Controls were divided into two groups, one set went inline and one set was not put in-line ("no-line"). The in-line controls were conducted using two chemical (organosilicone) dips, water only, 0.01 and 0.1% silicone defoamer combined with hot water spray temperatures of 10 and 27 °C and pressures at 0 and 400 kPa for both brush styles. The no-line controls were conducted using the same two chemical dips as the in-line sets, except they were returned directly to holding containers after dip treatment. There was a dry control, which stayed in the container and held at room temperature. Eight treatments were conducted using two chemical dips, 0.01 and 0.1% silicone defoamer in combination with the water spray temperatures set at 10 and 27 °C and pressures at 0 and 414 kPa. Each combination was tested for each brush style. After fruit were returned to the laboratory they were examined to determine the number of CM eggs remaining.

2.5. Test 4

This test was designed to determine the effects of defoamer concentration and water spray pressures on the removal of CM eggs from the fruit surface. Test combinations consisted of using hot water spray temperatures of 10 and 27 °C, pressures of 0, 200, 400, 550, 700, and 850 kPa with concentrations of 0.01 and 0.1% silicone defoamer. Washing brush style B was the only style used for this test.

Controls were divided into two groups, one set went inline and one set was not put in-line ("no-line"). The in-line controls were conducted using two chemical dips (water only, 0.01 and 0.1% silicone defoamer) combined with hot water spray temperatures of 10 and 27 °C and pressures at 0, 200, 400, 550, 700, and 850 kPa. The no-line controls were conducted using the same two chemical dips as the in-line set, but they were returned directly to holding containers after dip treatment. There was a dry control, which stayed in the container and held at room temperature. Treatments were conducted using two chemical dips (0.01 and 0.1% silicone defoamer) in combination with the hot water spray temperatures set at 10 and 27 °C and pressures at 0, 200, 400, 550, 700, and 850 kPa. After fruit were returned to the laboratory they were examined to determine the number of eggs remaining.

2.6. Statistical analysis

Statistical analyses were performed using SAS v. 8.0 (SAS 2003). Data were analyzed using factorial ANOVA and Duncan's Multiple Range Test. Regression was performed using QuattroPro (v.10.0).

3. Results

3.1. Test 1

The effects of 0.75% Silwet L-77 on egg hatch of the early and late stages of codling moth and oriental fruit moth eggs were negligible (Table 1). In the 600 s treatments, the percent hatch was higher than controls for the white ring of

Table 1 Percent egg hatch of codling moth and oriental fruit moth eggs as affected by immersion time in a solution of 0.75% Silwet

Stage	Control	Time (s)		
		60	300	600
Codling moth				
White ring	65.5 ± 3.1	64.4 ± 3.1	62.2 ± 3.1	69.5 ± 3.1
Black head	81.8 ± 4.4	85.6 ± 4.4	93.7 ± 4.4	90.4 ± 4.4
Oriental fruit me	oth			
White ring	69.4 ± 6.5	81.9 ± 6.5	71.9 ± 6.5	73.3 ± 6.5
Black head	92.6 ± 4.1	90.3 ± 4.1	83.2 ± 4.1	88.3 ± 4.1

Values not statistically significant from controls, ANOVA P > 0.05.

Table 2
Percent removal and egg hatch of mature codling moth eggs on apple as affected by Silwet L-77 concentrations, water pressure and water temperature sprays

Treatment			% Remove ^a	% Hatch
Temperature (°C)	kPa	Silwet		
Control	0	0	0 a	57.9 a
20	0	0	$36.2 \pm 3.6 \text{ b}$	$3.6 \pm 1.4 \text{ b}$
40	200	0	$69.2 \pm 4.3 \text{ c}$	$6.9 \pm 3.2 \text{ c}$
40	200	0.2	$66.9 \pm 3.8 \text{ c}$	$5.2 \pm 2.4 \text{ c}$
40	200	0.3	$75.2 \pm 1.7 \text{ c}$	$7.8 \pm 2.6 \text{ c}$
40	550	0	$91.1 \pm 2.6 d$	$0.5 \pm 0.5 \; d$
40	550	0.2	$89.6 \pm 3.1 d$	$0.5 \pm 0.5 \; d$
40	550	0.3	$91.9 \pm 4.9 d$	$0.0 \pm 0.0 d$
50	200	0	$85.7 \pm 2.0 \text{ c}$	$3.6 \pm 1.1 \text{ c}$
50	200	0.2	$75.5 \pm 4.0 \text{ c}$	$9.5 \pm 3.3 \text{ c}$
50	200	0.3	$82.0 \pm 3.3 \text{ c}$	$9.5 \pm 3.3 \text{ c}$
50	550	0	ND	ND
50	550	0.2	$96.2 \pm 2.1 d$	$0.0 \pm 0.0 \; \mathrm{d}$
50	550	0.3	$90.1 \pm 4.3 d$	$0.2 \pm 0.2 d$

^a Means with the same letters (a–d) in a column are not significantly different. Duncan's Multiple Range Test α = 0.05.

both CM and OFM and the black head stage of CM but were not statistically significant. Regression analyses resulted in R^2 values ≤ 0.5 , indicating no effect of dip duration on egg hatch.

3.2. Test 2

Approximately 60% of the eggs on the control fruit hatched (Table 2). This low hatch was probably due to fruit rotting. Over the length of time the fruit were held out at room temperature to allow for oviposition and egg development to the black head stage, approximately 7–10 d, the fruit began to show severe decay, which was noted in our evaluations. The 20 °C, 0 kPa also had poor egg hatch (3.6%), also an effect of fruit rot. Eggs from treatments using the 550 kPa water pressure showed the greatest percentage removal of eggs (89.6–96.2%) (P < 0.01, F = 52.77) and the lowest egg hatch (0.0-0.5%) (P<0.01, F=20.92). The addition of Silwet did not have a significant effect on either the removal of eggs or percent egg hatch (P=0.32,F = 1.5; P = 0.63, F = 0.45, respectively). The temperature of the water sprays did have a significant effect on egg removal and egg hatch only because the controls were included in the analyses (P < 0.01, F = 9.43; P < 0.01, F = 31.8, respectively). However, Duncans' multiple range test did not separate the means of the % removal and % hatch on the basis of the temperature in those samples that had been run over the

Neither the temperature of the spray water nor the presence of Silwet had a significant effect on the removal of ERM eggs from the fruit surface (Table 3). The most significant removal was achieved with the high-pressure sprays of 200 and 550 kPa (50–74% removal) (P<0.001; F = 32.94, df = 33) with no differences between the control and Silwet dips or the temperature of the spray water.

Table 3

Percent removal of European red mite eggs from pear fruit surfaces as affected by water pressure, spray water temperature, and Silwet L-77 concentration

Pressure (kPa)	Silwet (%)	Temperature (°C)	ERM ^a % removal \pm S.E.
0	No dip	20	$6.7 \pm 6.7 \text{ a}$
0	0	20	$13.0 \pm 6.7 \text{ a}$
0	0.3	20	$10.8 \pm 5.4 \text{ a}$
0	0	5.5	$15.8 \pm 8.0 \text{ a}$
0	0.3	5.5	$24.4 \pm 3.4 a$
200	0	30	$60.6 \pm 8.2 \mathrm{b}$
200	0	40	$64.1 \pm 12.1 \mathrm{b}$
200	0.3	30	$57.6 \pm 9.4 \mathrm{b}$
200	0.3	40	$58.2 \pm 11.0 \mathrm{b}$
550	0	30	$50.8 \pm 10.3 \mathrm{b}$
550	0	40	$61.7 \pm 18.4 \mathrm{b}$
550	0.3	30	$73.8 \pm 4.4 \mathrm{b}$
550	0.3	40	$58.9 \pm 10.0 \mathrm{b}$

^a Means with the same letters (a and b) in a column are not significantly different. Duncan's Multiple Range Test $\alpha = 0.05$.

3.3. Test 3

The high-pressure washing at $400 \,\mathrm{kPa}$ had a significant effect on the removal of CM eggs from the surface of the fruit, while the brush type and amount of defoamer did not add to the removal of the eggs from the fruit surface (Table 4). However, egg removal was significantly increased by the use of the brushes (P < 0.001; F = 12232.11, df = 118) (Table 4).

3.4. Test 4

The high-pressure sprays were more effective in removing CM eggs from the surface of the fruit (P < 0.001, F = 6.37, df = 4) (Table 5). Interestingly, in this test the packing line

Table 4 Effects of Defoamer, spray pressure, and brush type (A = firm, B = soft) on removal of mature codling moth eggs from apple surfaces

% Defoamer	Brush type	Water pressure (kPa)	% Removal \pm S.E.
0.01	0	0	$5.6 \pm 3.9 \text{ a}$
0.1	0	0	$19.1 \pm 4.7 \mathrm{b}$
0.0	0	0	$18.5 \pm 13.2 \mathrm{b}$
Dry	0	0	$20.1 \pm 6.0 \mathrm{b}$
0	A	0	$80.1 \pm 4.0 c$
0	В	0	$84.5 \pm 3.2 \mathrm{c}$
0	A	400	$90.7 \pm 1.2 \mathrm{c}$
0	В	400	$86.8 \pm 7.2 \text{ c}$
0.01	A	0	$63.0 \pm 13.7 \mathrm{d}$
0.01	В	0	$69.9 \pm 7.2 \mathrm{d}$
0.1	A	0	$76.9 \pm 8.1 \text{ d}$
0.1	В	0	$66.7 \pm 9.3 \mathrm{d}$
0.01	A	400	$92.0 \pm 2.5 c$
0.01	В	400	$81.9 \pm 6.6 \mathrm{c}$
0.1	A	400	$98.8 \pm 1.1 \text{ c}$
0.1	В	400	$89.9 \pm 1.0 \mathrm{c}$

Means with the same letters (a–d) in a column are not significantly different. Duncan's Multiple Range Test α = 0.05.

Table 5
Effects of defoamer (DF) concentration and spray water pressure on the removal of mature codling moth eggs from the apple surfaces

Pressure (kPa)	Removal ^a % ± S.E.	0.01% DF $\% \pm$ S.E.	0.1% DF % ± S.E.
0 _p	24.2 ± 7.0 a	30.2 ± 3.2 a	38.3 ± 1.9 a
0	$96.7 \pm 1.9 \mathrm{b}$	$73.4 \pm 4.6 \mathrm{b}$	$82.3 \pm 1.6 \mathrm{b}$
400	$99.1 \pm 0.9 \mathrm{b}$	$95.5 \pm 1.1 \mathrm{c}$	$98.4 \pm 1.1 \mathrm{b}$
550	$99.2\pm0.8\mathrm{b}$	$97.4 \pm 1.4 \mathrm{c}$	$100 \pm 0.0 \mathrm{b}$
700	$99.1 \pm 0.9 \mathrm{b}$	$98.1 \pm 1.3 \text{ c}$	$99.1 \pm 0.9 \mathrm{b}$
850	$100\pm0.0\mathrm{b}$	$100\pm0.0\mathrm{c}$	$99.1 \pm 0.9 \mathrm{b}$

^a Means with the same letters (a–c) in a column are not significantly different. Duncan's Multiple Range Test $\alpha = 0.05$.

control with the control dip removed the eggs as well as the pressurized sprays. Evidently there is an effect of the packing line which also aids in egg removal. The defoamers did not contribute significantly to the removal of eggs over that of the sprays. The most effective treatments, with egg removal greater than 95%, were obtained from spray pressures of 400 kPa and greater and the packing line control. The mere passage of the fruit over the packing line provided an average of 84% removal (Table 4).

4. Discussion

High water pressure appeared to have the highest impact on egg removal and reduction of egg hatch. The temperature of the water did not appear to have much of an impact on egg hatch or removal of either CM or ERM. This is in contrast to the findings of researchers in Israel (Fallik et al., 2000b), in which they claim that the high temperatures of the spray water had a significant effect on insect mortality. This may be due to the short duration our fruit are exposed to high temperature sprays, approximately 15 s as compared to approximately 25 s for the Israeli system (Fallik et al., 2000b).

Silwet and defoamers did not have a significant effect on reducing egg hatch or improving removal. Although it appears that Silwet is effective in removing and killing surface arthropods (Dentener and Peetz, 1992; Liu, 2000; Peetz and Dentener, 1992; Tipping et al., 2003; Wood, 1997), its effectiveness in removing or killing arthropod eggs has not been demonstrated in our research.

The high-pressure washing system is effective in removing CM and ERM eggs at pressures as low as 400 kPa, resulting in greater than 60% removal of ERM eggs and 90% of CM eggs. In situations where ERM egg levels exceed the 5% tolerance level in pre-packed fruit, the passage of these fruit across a high-pressure washing system will most likely bring these levels below the upper tolerance limits. To our knowledge, there is no restriction on the presence of CM or OFM eggs on fruit surfaces similar to those given for ERM eggs. However, there are restrictions on the existence of viable eggs and larvae of these pests. The use of high-pressure washing will increase fruit sanitation and reduce interception of these lepidopteran eggs regardless of viability.

Walker et al. (1999) found high-pressure washing to be very effective in removing California red scale from citrus fruit. They found that pressures of 1030–1380 kPa were as effective as water sprays at the recommended 2240 kPa. These pressures greatly exceed those used in our study, but no doubt the citrus, having a much denser cuticle, can withstand such pressures. We found that pressures 30% of those used in Walker et al. (1999) were sufficient to remove both ERM and CM eggs from the fruit surface.

Most other high-pressure washing references refer to decay control and commodity quality. We would like to emphasize that while we did see significant effects in the reduction of decay microorganisms with little effect on fruit quality, the effect of this system on the removal of surface arthropods and arthropod eggs are quite significant and can provide an additional level of pest control on the packing line. We believe that this system can play a significant role in a systems approach to pest control in commercially packed pears.

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^b Samples not run over packing line.

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